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Responses of skeletal muscle pH to injury: A new technique for determination of tissue viability

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Changes in hydrogen ion activity on the surface of organs and tissues are reliable experimental indicators of the degree to which metabolic activity in those tissues has shifted to the buildup of products of anaerobic metabolism.¹⁻⁴ Consequently, surface pH is logically assumed to be related to the adequacy of perfusion of the tissues being examined. In the intact kidney with occluded arterial inflow, surface hydrogen ion activity rises sharply and returns to normal after restoration of blood flow.³ In hemorrhagic shock, fall in surface pH of skeletal muscle may actually precede a measurable fall in blood pH or rise of lactic acid in the blood.⁴ Surface pH measurement may thus represent the most sensitive parameter by which adequacy of specific tissue perfusion may be assessed and closely monitored.

Since skeletal muscle accounts for a large portion of oxidative metabolism, it is reasonable that measurement of surface pH could have a valuable surgical application in assessment of limb viability following ischemia or trauma. A historic lesson of combat surgery is that the inexperienced surgeon often performs inadequate debridement because of the lack of objective evidence of nonviability of tissue. Furthermore, technically perfect arterial reconstructions have been performed in extremities with irrevers-

ible damage from ischemia. Over-enthusiastic, excessive debridement of viable tissue and limbs is likewise unacceptable.

Conventional criteria for estimation of viability of skeletal muscle include direct observation of color, consistency, contractility, bleeding, and the use of various staining methods. The latter techniques have definite limitations which render them impractical or imprecise for clinical application. The present study was designed to evaluate the technique of electrometric surface pH measurement in assessment of skeletal muscle viability following crush, thermal, and high velocity missile injury.

MATERIALS AND METHODS

The principles of laboratory animal care as promulgated by the National Society for Medical Research were observed. Twenty albino rabbits, weighing 2 to 3 kilograms, were anesthetized with intravenous sodium pentobarbital and methoxyflurane administered by the open-drop method. A 3 cm. incision was made through the skin and subcutaneous tissues of the lateral aspect of the thigh and deepened through the investing fascia, avoiding injury to muscle. Surface pH measurements were made on the underlying muscle with an Instrumentation Laboratories' Combination pH Glass Electrode Model 14043 held in light contact with the muscle surface and connected to an Instrumentation Laboratories Model 245 Delta-

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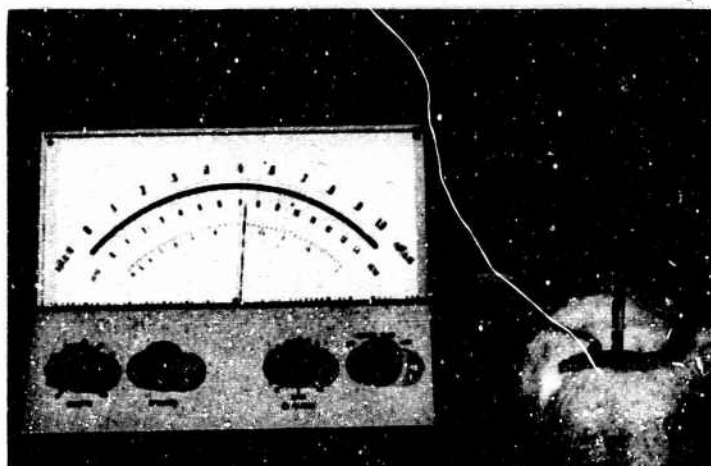


Fig. 1. Deltamatic pH/MV electrometer and electrode used for skeletal muscle surface pH measurements.

matic pH/MV Electrometer (Fig. 1). The instrument was calibrated with standard pH reference buffer solutions.

In 10 rabbits, crushing injury was created across the muscle of the lateral aspect of the thigh with a pair of Kocher clamps. The clamps were applied over a thin sheet of polyethylene to prevent contact of metal with muscle and left in place for 5 minutes. Serial pH determinations were made on the surface of normal and injured areas during a 2 hour period following injury. The injured area was then marked with metallic clips, the wound closed, and the animal returned to its cage. These wounds were reopened after 24 hours for a second measurement of surface pH. Then a correlation was made between this parameter and tissue staining after intravenous Patent Blue V dye.⁵

In 10 rabbits, a graded thermal injury was applied to the muscle by electrocautery with a Bovie Electrosurgical Unit, Model CSV. Three 1 cm. square areas of injury were created by varying the coagulation current in contact with muscle. Char injury was first produced with a current setting of 40 held in contact for 5 seconds. A lesser injury was then inflicted with a current setting of 25 to 30 for the same length of time. The setting was then reduced to 20 and held in contact with muscle for 10 seconds. These areas were arbitrarily designated as "severe," "moderate," and "mild" thermal

injury, respectively. Serial pH measurements were performed on the surface of each of the burned areas and over normal muscle. Results were then correlated with the degree of hypoperfusion indicated by tissue staining following intravenous Patent Blue V dye. After 24 hours, wounds were reopened and examined for gross and pH changes as well as staining characteristics accompanying vital dye infusion.

The effects of high velocity missile injury were studied in two beagle dogs. These animals were anesthetized with intravenous sodium pentobarbital and subjected to soft tissue wounding of each proximal hind limb with a 0.25 inch steel sphere traveling in excess of 2,000 feet per second. Animals were studied 2 to 3 hours following wounding. The entrance and exit wounds were fully exposed by incision and fasciotomy. Repeated skeletal muscle surface pH measurements were then made on multiple areas of traumatized muscle to determine differences in pH between normal tissue and muscle that usually would be excised in a similar combat wound occurring in man. Muscle debridement was not performed. Intravenous Patent Blue V dye was infused 4 hours after injury and again 24 hours later to correlate results of this parameter with gross changes and surface pH measurements. Results of all pH determinations were subjected to statistical analysis, and probability was determined by Student's nonpaired t test.

Table I. Skeletal muscle surface pH measurement accompanying crush, thermal, and high velocity injury

Description	Mean	Range	S.E.*	Significance
<i>Crush injury</i>				
Normal	7.47 ^o	7.358-7.610	0.025	p < 0.001
Crush	6.585	6.334-6.875	0.058	
<i>Thermal injury</i>				
Normal	7.557	7.340-7.722	0.046	N.S.
Mild	7.631	7.443-7.742	0.055	
Moderate				
Immediate	6.914	6.600-7.241	0.066	p < 0.001
30 minutes	7.449	7.165-7.584	0.050	p < 0.001
Severe	7.652	7.205-7.828	0.110	N.S.
<i>High velocity</i>				
Normal	7.786	7.568-8.065	0.025	p < 0.001
Injured	7.280	7.178-7.448	0.041	

*S.E. = standard error.

RESULTS

The results of surface pH measurement of skeletal muscle subjected to crush, thermal, and high velocity missile injury are expressed in Table I and Figs. 2 and 3. Immediately following crush injury, the muscle was white and obviously totally ischemic, and the surface pH varied minimally from normal. However, fall in pH was progressive during the 30 minutes following injury. During this time, traumatized muscle became more reddish in color, as blood flow to the injured area was restored. pH of injured skeletal muscle was stable by 1 hour after injury, with mean pH fall of 0.894 ± 0.065 pH units ($p < 0.001$). Surface pH of adjacent normal muscle remained stable during this period with transition from normal to acid pH values regularly occurring at the point of gross demarcation of the crush injury. On the day following injury, pH also changed sharply at the point of demarcation outlined by the metallic clips and by tissue staining after intravenous Patent Blue V dye (Fig. 4). At this time, however, pH change was in the opposite direction. Surface pH of crushed tissue was always higher than that of surrounding normal tissue, still with the sharp line of demarcation observed on the day of injury.

There was no detectable fall in surface pH of skeletal muscle subjected to "mild"

thermal injury. However, surface pH of skeletal muscle that was burned more severely ("moderate") regularly demonstrated a transient significant pH fall, followed by restoration to normal levels within 30 minutes following injury (Fig. 2). Mean fall in pH in moderately burned areas was 0.655 ± 0.078 ($p < 0.001$). Restoration to normal values after 30 minutes was also statistically significant ($p < 0.001$). Intravenous administration of Patent Blue V dye after burning caused staining in minimally burned areas but initial failure of staining in areas subjected to moderate injury. However, the latter areas subsequently became bright blue, concomitant with rise in surface pH values toward normal. Areas of most severe injury (char) were not stained by the dye, and pH did not fall. Twenty-four hours later charred tissue was usually alkalotic.

High velocity missile injury with the symmetrical steel sphere seemed to cause less gross tissue destruction than that occurring after injury with an M-16 rifle, a high velocity weapon currently used in combat areas. The usual small wound of entrance and larger exit wound were nevertheless created, with areas of muscle appearing definitely devitalized by conventional clinical criteria. Surface pH of traumatized muscle was consistently lower than normal muscle. Average pH fall in apparently devitalized

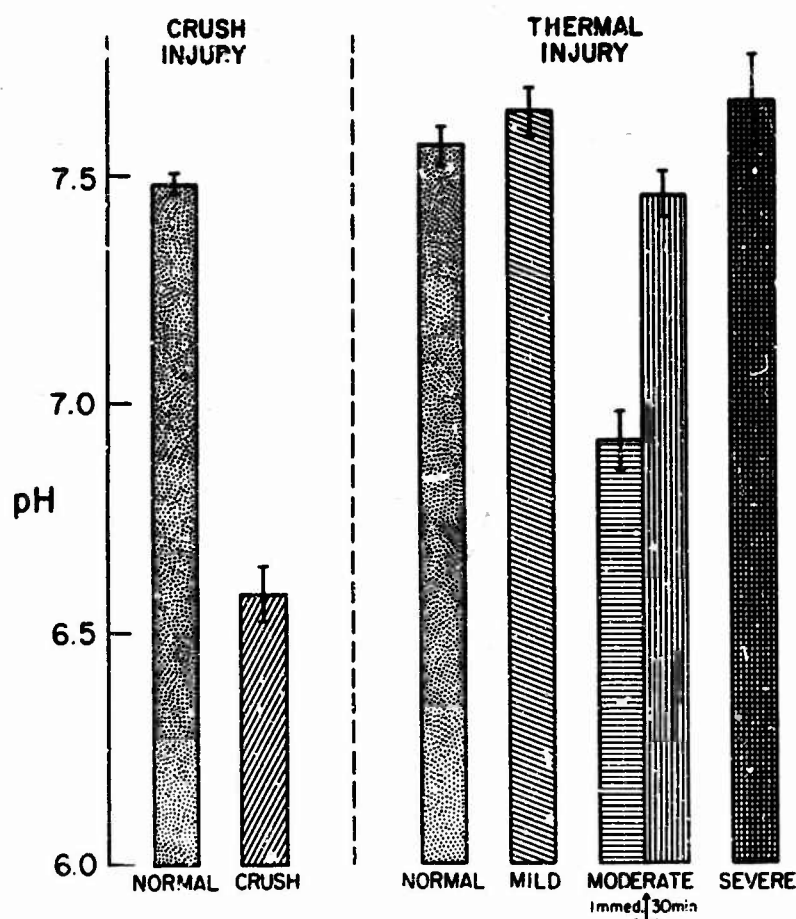


Fig. 2. Skeletal muscle surface pH following crush and thermal injury. Values expressed represent the mean of all determinations \pm standard error.

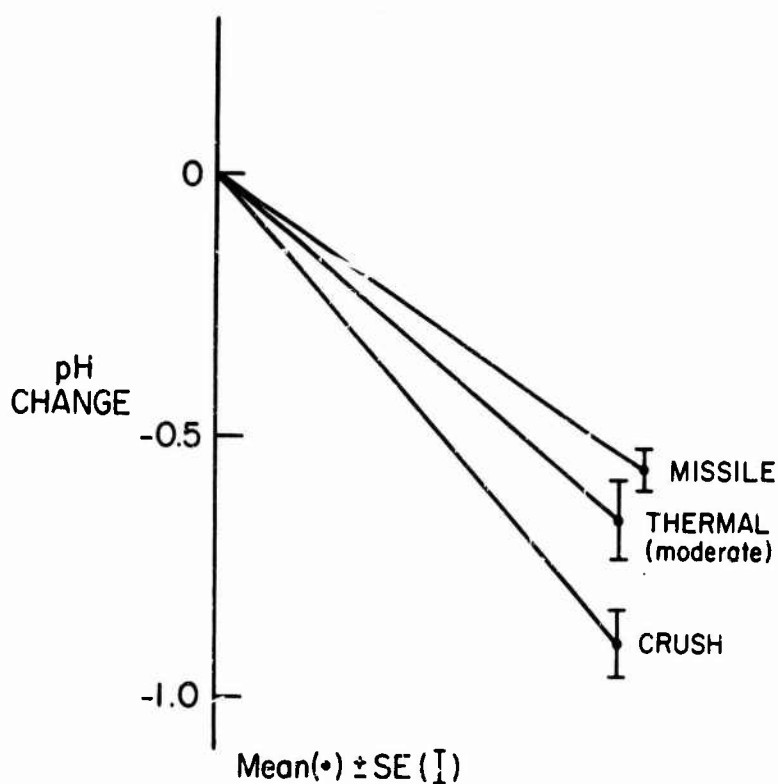


Fig. 3. Skeletal muscle surface pH changes following crush, burn, and missile injury. Values expressed represent the mean of all determinations \pm standard error.

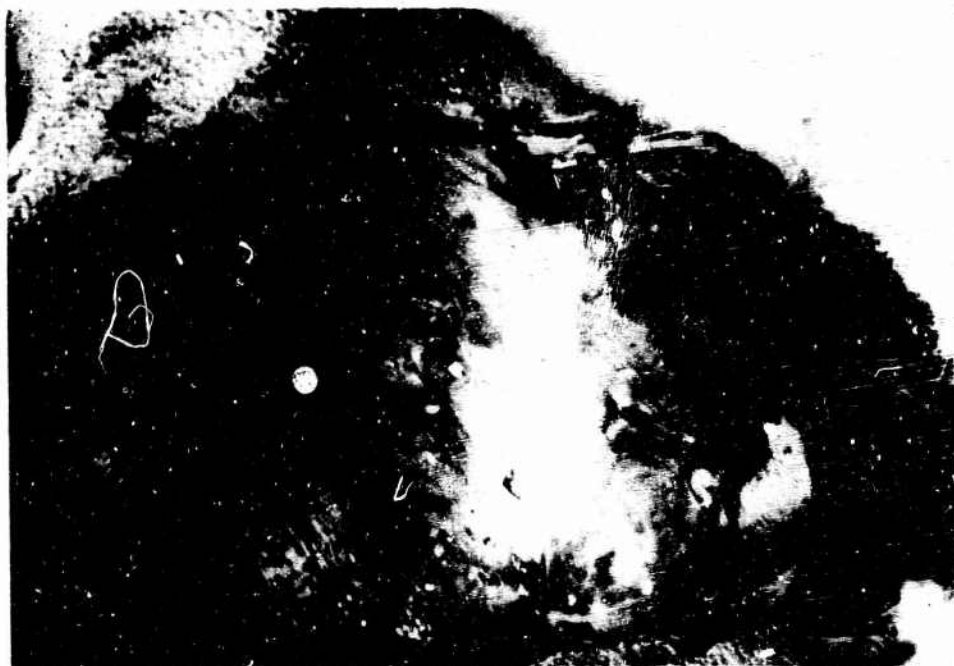


Fig. 4. Staining of tissues accompanying Patent Blue V dye administered intravenously after crush injury. The unstained area (arrows) corresponds to area of crush.

areas was 0.506 ± 0.037 pH units (Fig. 3). Muscle staining after infusion of Patent Blue V dye was poor on the day of injury, except in one wound in which correlation of staining with grossly devitalized muscle seemed reliable. Twenty-four hours later, however, demarcation by gross observation, vital dye infusion, and fall in muscle surface pH were all possible. The shift to alkaline pH of apparently devitalized muscle which was observed 24 hours after crush and severe burn injury had not occurred after missile injury.

DISCUSSION

Diminution of surface pH of skeletal muscle after wounding suggests that cellular metabolic activity in traumatized muscle readily shifts to the accumulation of products of anaerobic glycolysis. The extent of this transition in the acute period after injury would appear to be directly related to the extent of tissue devitalization and local accumulation of lactic acid and other acid metabolites.^{1, 2}

Correlation of surface pH with intravenous infusion of vital dyes indicates that changes in surface hydrogen ion activity are

directly related to alterations in tissue perfusion. The absolute fall in pH of muscle injured by crush or high velocity missile and the transient fall in pH in tissue subjected to moderate thermal injury reflect such alterations in perfusion. In moderately burned areas, correlation of the restoration of pH to normal values with the appearance of staining after vital dye infusion further indicates that tissue perfusion was restored toward normal by 30 minutes following injury. The slight rise in surface pH in tissue subjected to minimal burn may be the reflection of augmented tissue perfusion. Although consistent, this change is not statistically significant.

Failure of charred muscle to demonstrate a fall in pH is believed to reflect absolute tissue devitalization with cellular death and total metabolic failure. After 24 hours, charred areas, as well as those subjected to crush injury, regularly demonstrated an increase in surface pH values above those in normal muscle, with distinct lines of demarcation from normal muscle. However, this phenomenon was not observed 24 hours after high velocity missile injury. It is likely that the total devitalization that followed

crush and char injury probably did not occur with the less uniform though equally severe injury inflicted by a high velocity missile. It would thus appear that tissues absolutely deprived of blood supply do not demonstrate the same capacity for maintaining surface acidosis as those in which some measure of perfusion is maintained. It is not clear from these studies whether this represents diminished capacity for anaerobic glycolysis² or loss of integrity of the cell membrane.

Demonstration of changes in surface hydrogen ion activity of injured muscle suggests that electrometric measurement of surface pH may be useful in assessing and monitoring tissue viability in cases of severe peripheral vascular insufficiency and direct trauma. Such a clinical adjunct could be of special value in the following instances: for wounds of critical areas in which extensive debridement might result in unnecessary alteration of appearance or function (e.g., face, hand), for areas in which viable muscle could better protect another surgical procedure (e.g., femoral vascular repair), and for difficult areas in which massive trauma is accompanied by a high incidence of re-debridement and/or infection (e.g., pelvis, retroperitoneum). Since equilibration of electrode and muscle surface is not usually instantaneous, however, direct application to combat surgery may be somewhat cumbersome with currently available instrumentation.

The results of this study would suggest that significant devitalization has occurred after injury when repeated surface pH determinations demonstrate significant variation from obviously normal tissue. When this difference exists and when the tissue being studied is acidotic, it is likely that impairment of perfusion has been recent. When the tissue in question is significantly alkalotic relative to obviously normal muscle, absolute devascularization or death has probably oc-

curred considerably earlier. Theoretically, these findings would render the techniques of surface electrometric measurement most applicable in the acute period following injury or vascular insult when precise definition of viability is most difficult. Additional studies are now in progress to correlate the extent of change in surface pH with ultimate tissue death or survival following high velocity missile injury and vascular occlusive disease.

SUMMARY AND CONCLUSIONS

Surface electrometric pH of skeletal muscle demonstrates significant changes after crush, thermal, and high velocity missile injury. The anatomic correlation of these changes with the distribution of vital dye indicates that surface hydrogen ion activity of skeletal muscle is readily altered by changes in tissue perfusion. Further studies will be directed toward definition of the extent of pH change which is consistently associated with tissue death.

REFERENCES

1. Couch, N. P., Maginn, R. R., Middleton, M. K., Appleton, D. R., and Dmochowski, J. R.: Effect of ischemic intervals and temperature on renal surface hydrogen ion concentration, *Surg. Gynec. & Obst.* 125: 521, 1967.
2. Couch, N. P., and Middleton, M. K.: Effect of storage temperature on the electrometric surface hydrogen ion activity of ischemic liver and heart, *SURGERY* 64: 1099, 1968.
3. Dmochowski, J. R., Couch, N. P., Kempf, R. N., and Appleton, D. R.: Electrometric surface pH of the ischemic kidney and the effect of hypothermia, *J. S. Res.* 6: 45, 1968.
4. Lemieux, M. D., Smith, R. N., and Couch, N. P.: Effect of hemorrhage and infusion on electrometric surface hydrogen ion activity and redox potential of skeletal muscle, *S. Forum* 19: 1, 1968.
5. Randolph, J. G., Leape, L. L., and Gross, R. E.: The early surgical treatment of burns. I. Experimental studies utilizing intravenous vital dye for determining the degree of injury, *SURGERY* 56: 193, 1964.